

ADDITIONAL METHODS FOR THE 1997 MONITORING YEAR

Springsnails still have yet to recolonize Site 1 (Hot Creek) since a major runoff event in July 1992 (Mladenka 1992, Varricchione and Minshall 1997). The following environmental and biological measurements/experiments were made/conducted in 1997 to gain greater information on the ecology of *P. bruneauensis* and to determine which factors have been preventing its recolonization of Hot Creek. Also, additional information was collected on the environmental conditions of the rockface seep sites (2, 3-OS, and 3-NS).

Environmental Measurements

Further analysis of substrate quality in Hot Creek (Site 1).

The Hot Creek streambed has become dominated by sand, silt, and small gravel as a result of grazing activity and flooding events that occurred in the early 1990's (Mladenka 1992, Varricchione and Minshall 1997). Large substrate is thought to be the best Springsnail habitat because it provides surfaces conducive to snail egg-laying (Mladenka 1992). Three trenches (0.50 m long x 0.25 m wide), spaced 50 m apart, were excavated in Hot Creek on 21 July 1997 to determine if there was a large amount of gravel and cobble substrate beneath the fine surficial sediments.

Discharge monitoring at the rockface seeps.

The water emerging from these seeps is diffuse, making it difficult to monitor flow. Small 90° V-notch weirs were installed approximately 1 m from the rockface seeps on 17 October 1997. The weirs collected diffuse runoff coming from the rockface to permit estimation of spring-flow discharge. The approximate location of the weirs is shown in Figure 2.

Rockface habitat mapping at Sites 2, 3-OS, and 3-NS.

Springsnail habitat area and quality were described for the

entire rockface seep areas at Sites 2, 3-OS, and 3-NS on 15 November 1997. These measurements were made in conjunction with total snail population counts (for description see below).

Biological Measurements and Experiments

Total Springsnail counts at the rockface study sites.

The current monitoring methodology does not allow for the determination of total numbers of snails present at a given site or within different habitat types on the rockface seep habitats. Therefore, an additional survey method was added to the protocol to produce estimates of total Springsnail numbers on an annual basis.

At each site, the entire rockface seep area was divided up into approximately 0.5 x 0.5 m subunits using a tape measure as a baseline across the rockface. A 8.5-cm diameter ring was placed in the center of each subunit and the snails within the ring were counted. Also, the habitat type (springflow conditions and relative cover by thick, orange periphyton complex) was estimated for each subunit.

Intensive search for relict populations of P. bruneauensis in and around Hot Creek.

Since *P. bruneauensis* has not been found at the Hot Creek study site for the past several years (Varricchione and Minshall 1997, 1996, 1995a; Royer and Minshall 1993), it is important to determine if potential recolonists for Site 1 occur anywhere in, or adjacent to, the stream between Indian Bathtub and the Bruneau River. Robinson and others (1992) had described a small stream-side refugium that had retained < 10 Springsnails after flooding and scouring events in the same year. As grazing pressure was lifted from the Hot Creek area, the growth of thick riparian vegetation near the creek and the seep made observation of this population difficult (Royer and Minshall 1993, Varricchione and Minshall 1997). An intensive search for relict populations of *P. bruneauensis* was conducted on 21 July 1997 in and immediately adjacent to Hot Creek (between Indian Bathtub and the Bruneau

River). The search was completed by examining (without magnification) Hot Creek sediments, emergent vegetation, and nearby rockface seeps for *P. bruneauensis*. Where Springsnails were found, temperatures were recorded using a Reotemp digital thermometer (model TM99A).

Controlled fish-feeding experiment in Hot Creek.

Populations of fish, redbelly tilapia (*Tilapia zillii*) and mosquito fish (*Gambusia affinis*), may be responsible for the disappearance and/or lack of recovery of the Bruneau Springsnail populations (Varricchione and Minshall 1997). These fish have been stocked worldwide, including the Great Basin ecoregion (Sigler and Sigler 1987). *T. zillii*, has a diet that is most often dominated by aquatic macrophytes, although it is believed to be omnivorous (i.e. feeding also on algae, invertebrates, and other fish). *G. affinis*, is known for its predation on mosquito larvae, but it also is known to eat a wide range of other food items (e.g. diatoms and other algae, crustaceans, and insects (Sigler and Sigler 1987)). *G. affinis* also has been documented for its cannibalistic behavior (adult consumption of juveniles) (Dionne 1985). Even if these fish do prey on *P. bruneauensis*, the operculum and shell of the Springsnails may act as a barrier to digestion in the fish gut (Norton 1988). Fish feeding on Springsnails in other locations within the Bruneau River drainage might even act as a dispersal mechanism.

Previous analysis of the gut contents of several *Tilapia* and *Gambusia* revealed no Springsnails (Varricchione and Minshall 1995b), but Springsnail populations had not been evident in Hot Creek at the time. A controlled fish-feeding experiment was conducted in 1997 to determine the extent to which the local fish might eat the Springsnails, and whether the snails can survive the process. Stomach content analysis was employed to evaluate this fish-feeding impact (Varricchione and Minshall 1995b, Dionne 1985, Gregory and Northcote 1993).

Blue plastic containers (47 cm x 34 cm), with the sides removed and replaced with 53- μ m mesh to permit the flow of water, were used as experimental enclosures. The enclosures were

secured in Hot Creek (approx. 20 m upstream from the confluence of Hot Creek and the Bruneau River) with steel rods. One hundred Springsnails were collected from Site 2 and brought to the experiment location. Twenty-five Springsnails were placed in each of the 4 enclosures. In each enclosure, Springsnail composition was 5 snails which were approximately 2.10 mm and 20 snails which were approximately 0.91 mm. The first 2 enclosures contained 3 *T. zilli* each (10.33 ± 0.45 cm and 10.17 ± 0.21 in the "Springsnail-only" and "Springsnail-plus-additional-food" treatments, respectively). The second 2 enclosures contained 3 *G. affinis* each (2.78 ± 0.36 cm and 2.70 ± 0.37 cm in the "Springsnail-only" and "Springsnail-plus-additional-food" treatments, respectively). The fish had been trapped using dipnets. In one of the *T. zilli* treatments and one of the *G. affinis* treatments additional food choices were provided because these fish are believed to be omnivorous (Sigler and Sigler 1987). The additional food consisted of (approximately): 3 20-cm stream biofilm-colonized sedge stalks, 3 stream biofilm-colonized *Salix* sp. leaves, 20 g of detritus (primarily grasses), 10 adult beetles (*Cleptelmis* sp.), 2 algae-colonized rocks, and 1 10-cm macrophyte clipping (*Elodea* sp.).

The fish and potential food sources were placed into their proper enclosures at 15:30 on 14 November 1997. Upon returning at 17:30 on 15 November 1997, the enclosures were removed of their contents, which were then placed on ice. In the laboratory, the samples were kept frozen until processed. During processing, the stomachs of the fish were removed by dissection and examined under a Bausch and Lomb dissecting microscope at 7x magnification. The occurrence of organic matter and/or invertebrates (including *P. bruneauensis*) was recorded.

Movement rates of *P. bruneauensis* under different food resource availability conditions.

In order to determine whether the range of movement of Springsnails hindered their ability to naturally recolonize habitats from which they have been extirpated, a Springsnail movement-rate experiment was conducted in Hot Creek. To determine these rates, plastic containers (25 cm x 14 cm x 7 cm),

with the sides removed and replaced with 53- μ m mesh to permit the flow of water, were used as experimental enclosures. The three enclosures contained 36 ceramic tiles (each 2.3 cm x 2.3 cm) positioned in a rectangular grid of 9 rows and 4 columns. In the first enclosure the tiles were completely devoid of algae. The second enclosure contained tiles that had been previously colonized by algae in Hot Creek for one month. These tiles were then gently scrubbed with a toothbrush to remove approximately half of the algae. Tiles in the third enclosure were colonized for one month and left undisturbed. Chlorophyll a and AFDM measurements were made on one tile for each of the algae treatments. Springsnails, taken from Site 2 and approximately 1.14 mm in size, were used in the experiments. Twenty-five snails were placed in each enclosure. The snails were placed on the downstream, left-most tile of the enclosures at the beginning of the experiment. Springsnail locations in the enclosures were recorded at 14:30 and 16:00 on 14 November 1997 and at 12:00 and 18:00 on 15 November 1997.

RESULTS

Springsnail Size Distribution

From 1990 to 1993, snail size distribution was monitored at three study sites: Site 1 (Hot Creek), Site 2 (upper spring rockface), and Site 3-OS (lower spring rockface) (Mladenka 1992). As suggested by Royer and Minshall (1993), a new seep at the southern edge of Site 3-NS was included in Springsnail monitoring for 1994 through 1997.

Site 1 (Hot Creek)

Site 1 (Hot Creek) population density was reduced to nearly zero in July 1992 and had yet to recover, as of November 1997 (Figs. 3h, 7). The flood in July 1992 probably resulted in the death of younger snails and skewed the size distributions in July and September 1992 (Fig. 3c). Mean size distribution data suggest that when the Springsnails were present (1990-1992), life histories were correlated with season, and a single cohort of individuals moved from juvenile classes in the winter to mature classes in the summer (Fig. 5a).

Site 2 (Upper Spring Rockface)

The Springsnail population at Site 2 maintained a size distribution that was relatively even across size classes between February and November 1997 (Fig. 3h). This trend agreed with monitoring results from previous years (Figs. 3a-g). Mean size distribution data (Fig. 5a) showed juveniles to be prevalent at all times of the year.

Site 3-OS (Lower Spring Rockface)

There were no clear size distribution trends between February and November 1997, although the warmer months (August and September) did have a larger proportion of individuals that were ≥ 1.5 mm, either being indicative of the growth of a cohort between the spring and summer months or outlier data (Fig. 3h).

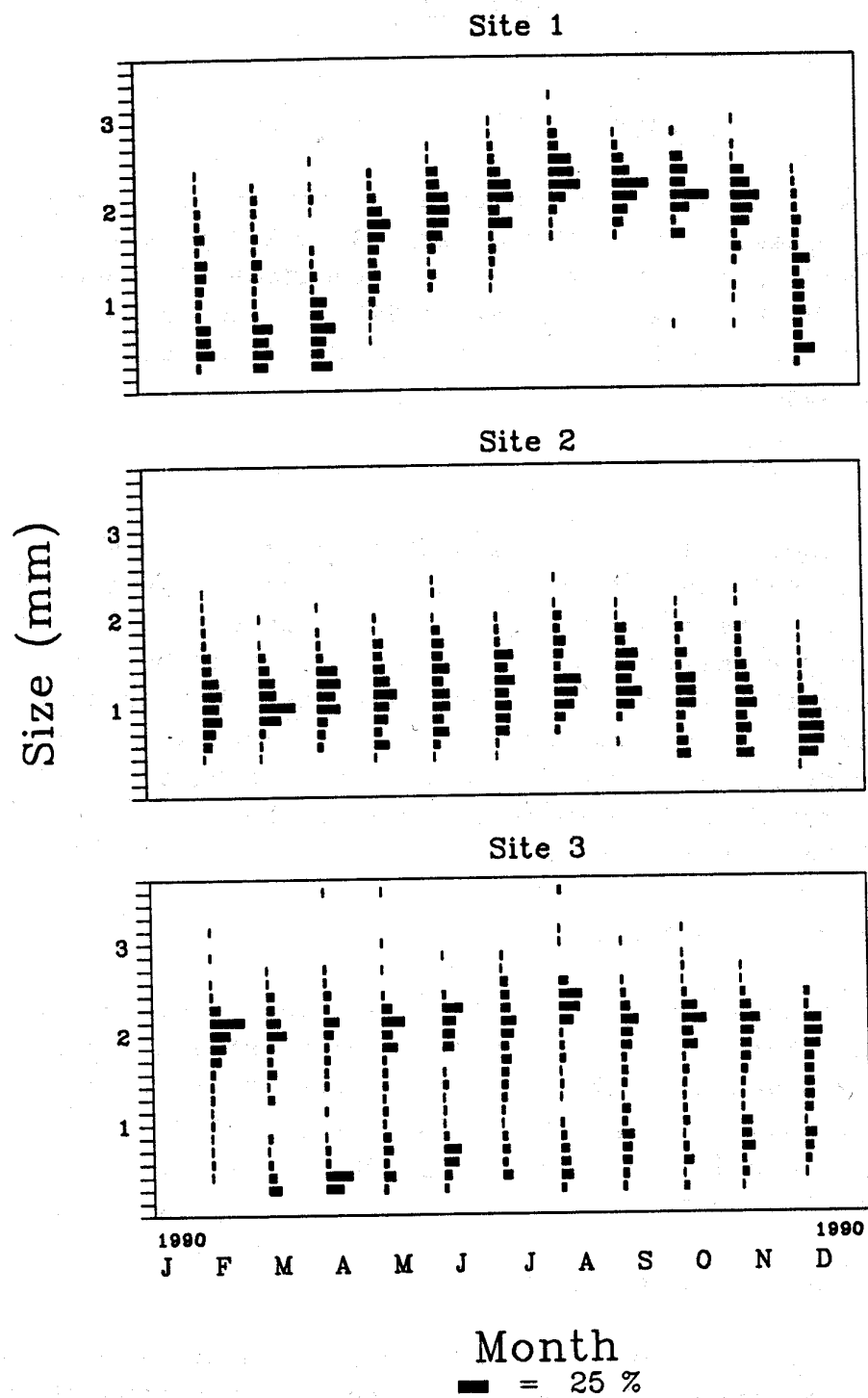


Figure 3a. Size histograms for the Bruneau Springsnail study sites. Horizontal tick marks represent 0.14mm size classes. Solid bars represent relative abundance of snails for a particular size class (n=100 for each sample).

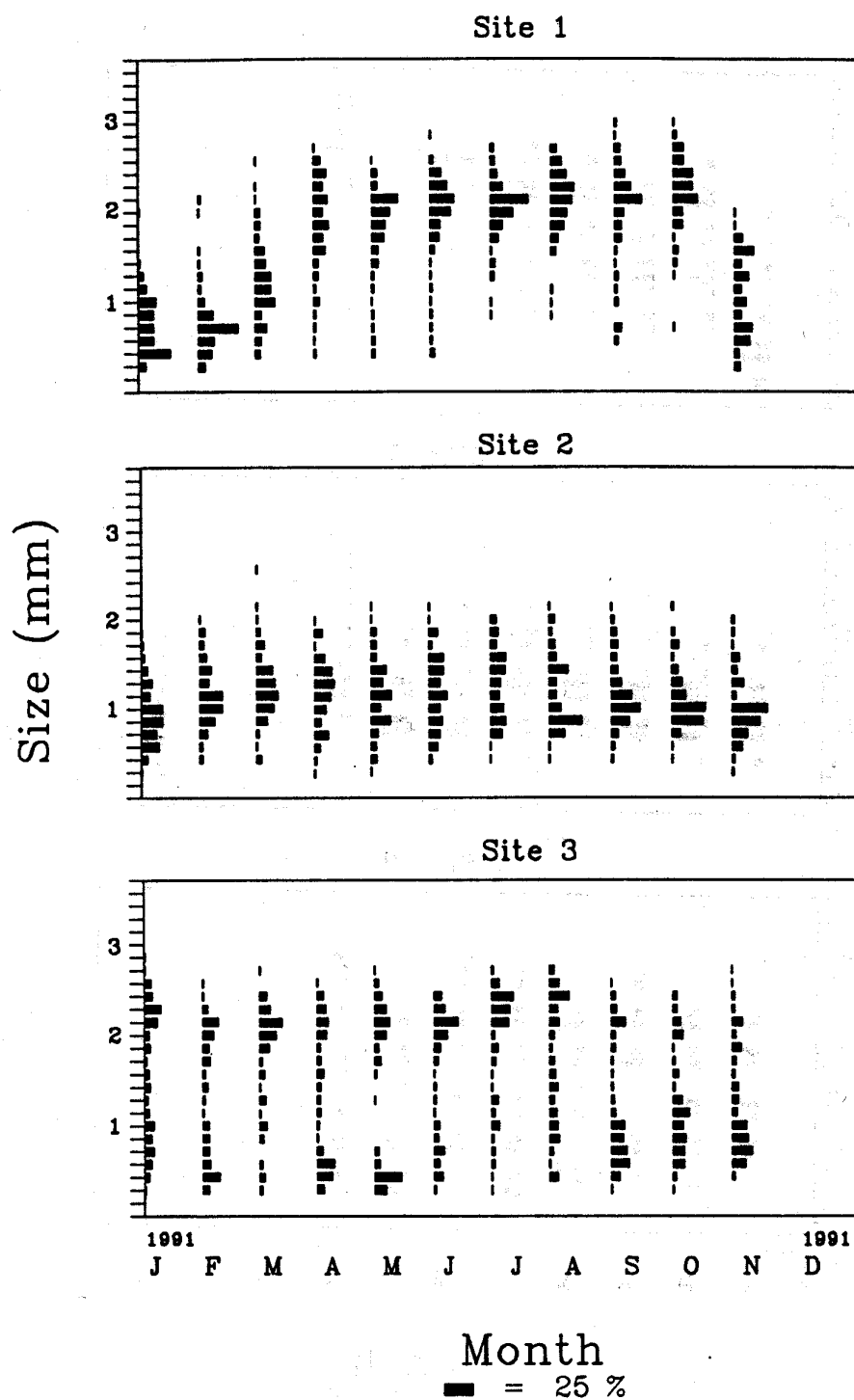


Figure 3b. Size histograms for the Bruneau Springsnail study sites. Horizontal tick marks represent 0.14mm size classes. Solid bars represent relative abundance of snails for a particular size class (n=100 for each sample).

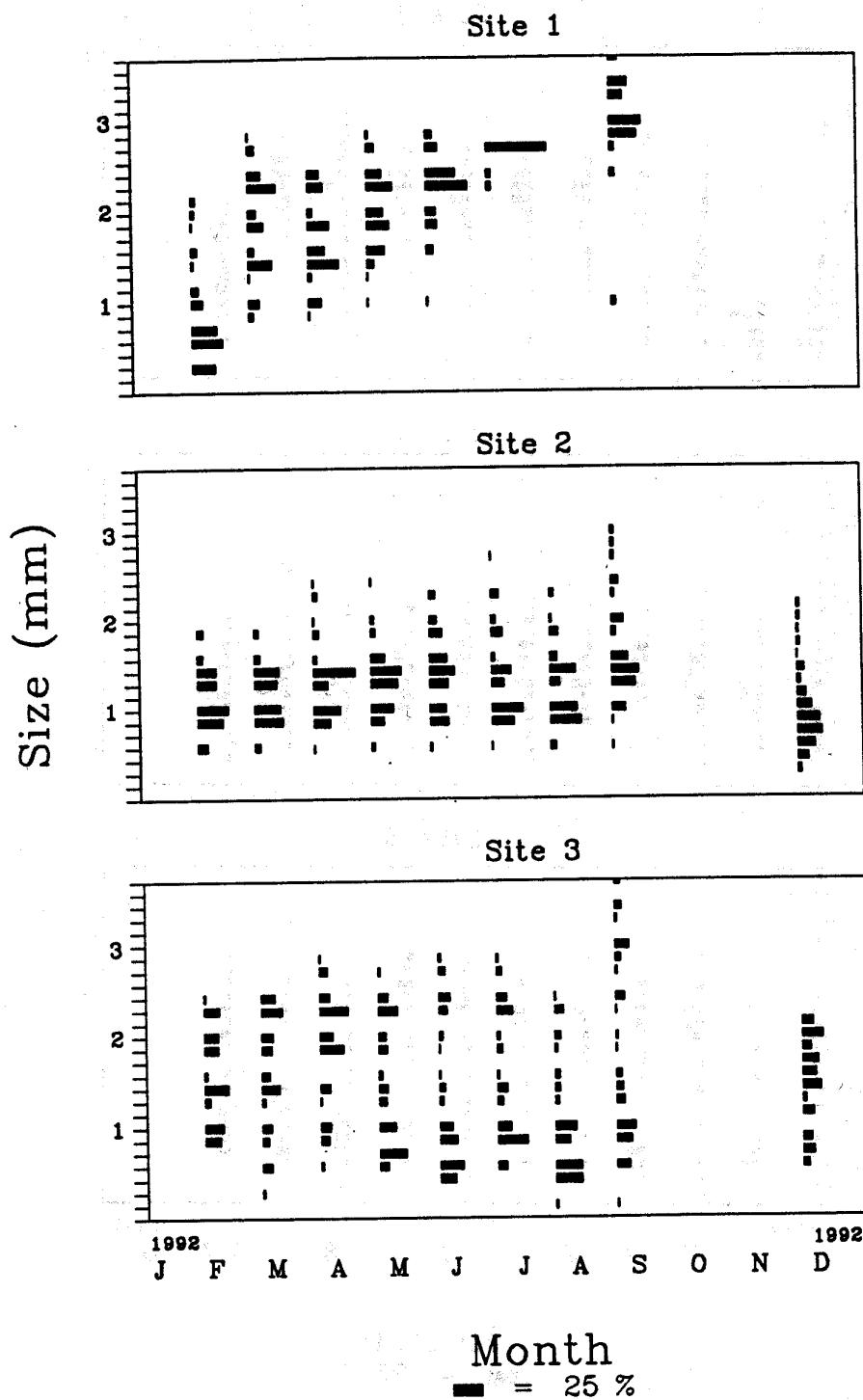


Figure 3c. Size histograms for the Bruneau Springsnail study sites. Horizontal tick marks represent 0.14mm size classes. Solid bars represent relative abundance of snails for a particular size class (n=100 for each sample). In July, 92% of the snails at Site 1 were found in the 2.66 mm size class (an out of range value for this figure).